



# Upholding Seed Quality with Mid-storage Invigoration Treatments in Rice (*Oryza sativa* L.)

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## Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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## ABSTRACT

The groundbreaking research investigated the impact of mid-storage invigoration treatments on seed quality parameters in rice. The experimental design used in the study was Completely Randomized Design with three replications. The experiment was conducted at the Regional Agricultural Research Station, Pattambi, Kerala Agricultural University. The objective of the study was to understand the effect of mid-storage invigoration treatments on seed storage under ambient conditions. An experiment was conducted to evaluate the effect of ten different treatments on the quality of seeds, specifically the germination percentage, seed vigour index I, electrical conductivity, and mean germination time. The treatments tested were control- untreated seeds, hydration and

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dehydration treatment 1l/kg, ascorbic acid 10g/kg, calcium chloride 10g/kg, albizzia leaf powder 2g/kg, neem cake 5g/kg, neem leaf powder 2g/kg, neem oil 2ml/kg, red chilli powder 1g/kg and vayambu leaf powder 2g/kg. The rice variety Jyothi was used. Results showed that all the mid-storage invigoration treatments significantly improved the germination rate of the treated seeds when compared to the control under ambient storage conditions. Neem oil when used showed the highest germination percentage after five months of storage. In the last month of storage, neem oil showed a significantly higher vigour index which was comparable to hydration dehydration treatment and red chilli powder. Germination percentage and seedling vigour index I are positively associated with seed quality. Electrical conductivity and mean germination time are negatively correlated with seed quality. The least electrical conductivity was recorded in hydration dehydration treatment. Neem oil and hydration dehydration treatment had low mean germination time. Based on the research conducted, neem oil, hydration-dehydration, and red chili powder are the most effective treatments for improving the longevity of seeds. These eco-friendly and cost-effective mid-storage invigoration seed treatments can help farmers enhance the quality of their seeds and extend their storage life.

*Keywords: Midstorage invigoration; germination %; seedling vigour index I; electrical conductivity; mean germination time.*

## 1. INTRODUCTION

Rice production is anticipated to increase globally, stressing its importance in tackling food security issues [1]. The availability of high-quality seeds is critical to ensuring a bountiful harvest and sustainable agriculture. High-quality seeds lead to better germination rates, resulting in healthier seedlings. Improved germination directly translates to higher yield potential for paddy crops. Farmers who use viable seeds can expect better productivity compared to those using subpar seeds. It is essential to emphasize the economic significance of viable paddy seeds for small-scale farmers and support their efforts to obtain the best possible seeds for their farming operations [2]. Therefore, it is imperative to ensure that farmers have access to high-quality seeds and that they are stored and distributed correctly. With the right seeds, farmers can improve their yields and income, contributing to food security and economic development [3].

Rice cultivation by small-scale farmers plays a significant role as it ensures food security, provides essential nutrients and generates income. The practice of preserving, utilizing, and exchanging viable seeds is prevalent for subsistence among these farmers. Prolonged seed storage significantly enhances this essential activity, ensuring a sustainable supply of seeds for subsequent seasons. Small-scale farmers in Kerala face the challenge of preserving paddy seeds for extended periods to ensure successful crop cycles. Farmers can consider storing seeds in ambient storage, which is a cool, dry, and well-ventilated room that can

preserve seed quality for up to six months. Long-term storage plays a critical role in maintaining seed quality and minimizing the need for frequent seed purchases. As the seed is hygroscopic in nature, its quality is being affected due to variations in the environmental conditions viz., relative humidity, temperature, moisture content, gaseous exchange, packaging material etc [4]. A practical solution is needed to tackle this problem and ensure the quality of the seeds.

Simple and affordable technologies are more suitable for small scale farmers involved in seed storage to ensure viable seed input for succeeding season. Adapting mid-storage invigoration treatments is a highly effective solution that can help maximize the quality and longevity of valuable seed assets. The cost-effective treatments are effective choices for this purpose. This act as a cornerstone in preservation and protection of upgraded seeds in seed storage. Several studies have demonstrated the efficacy of these mid-storage treatments for paddy seeds. A study [5] found that neem oil treatment significantly improved the seed quality parameters of paddy seeds during storage. Similarly, it was reported [6] that hydration and dehydration treatment helped in maintaining the moisture content of paddy seeds, preventing their deterioration during storage. Additionally, it was demonstrated [7] the effectiveness of red chili powder in controlling pests and fungal growth in stored paddy seeds.

During storage, midstorage invigoration treatments changes induce changes in seed quality parameters such as germination

percentage, seedling vigor index I, mean germination time and electrical conductivity which plays a vital role in preserving seed viability. Germination percentage measures the proportion of viable seeds in a given sample, while seed vigor index indicates the overall seed quality by measuring the seedling growth rate and vigor. These parameters are useful for predicting seed longevity and quality, as well as for identifying seed lots that require immediate use or disposal. Several studies have shown the importance of seed quality parameters during seed storage. In a study [8], it was found that the germination percentage of seeds decreased significantly with increasing storage time, indicating a decline in seed viability. Similarly, it was reported [9] that seed vigor index was a better predictor of seedling emergence than germination percentage, highlighting its importance in assessing seed quality during storage. Seed viability and vigor decrease over time during storage, leading to reduced seed germination and establishment. These findings emphasize the need for careful monitoring of seed quality parameters during seed storage to maintain seed viability and ensure successful crop production.

Other important factors that affect seed viability during storage are electrical conductivity and mean germination time. Electrical conductivity is an indicator of seed deterioration and is associated with the leakage of electrolytes from damaged cells. High electrical conductivity values indicate poor seed quality while low values are indicative of good quality seeds [10]. The electrical conductivity of seeds increases with damage to the cell membranes and loss of membrane integrity, leading to reduced seed quality and viability. Therefore, reducing electrical conductivity is essential to ensure long-term storage and conservation of plant genetic resources. Several studies have highlighted the importance of reducing electrical conductivity and mean germination time during seed storage. A study [11] found that reducing electrical conductivity during storage improved seed quality and enhanced the performance of seeds. Mean germination time, on the other hand, measures the average time taken for seeds to germinate under optimal conditions, and is used as an indicator of seed vigor. The longer the mean germination time, the lower the seed vigor and viability. Therefore, a lower mean germination time is desirable to ensure high seed quality and long-term storage [12].

Various compounds and techniques are used for seed treatments in midway through storage to invigorate the seeds and are efficient in preserving high viability and vigour of seeds. However, there is a need to identify cost effective and eco-friendly midstorage invigoration treatments which are easily adoptable by small scale farmers. In this context this article explores midstorage invigoration treatment interventions in paddy seed storage focusing on the popular variety Jyothi. By investigating the efficacy of these treatments, the study aims to provide insights into the most effective as well as farmer friendly treatments that enhance seed quality and storage period contributing to higher seed production and productivity.

## 2. MATERIALS AND METHODS

### 2.1 Experimental Design

Ten treatments were applied to paddy seeds of the Jyothi variety at six months of age. Seed quality parameters like germination percentage, seedling vigor index (I), electrical conductivity, and mean germination time were measured before and after treatments at monthly intervals. The observations were recorded until the seed germination rate dropped to 80%. A Completely Randomized Design (CRD) was used for the analysis of seed quality parameters. For every treatment, three replications were examined, with a quarter of a kilogram of seeds per replication. After that, the seeds were placed in gunny bags and left to ambient conditions. Experiment ran from October 2022 to March 2023, using seeds from both treatments and control groups.

### 2.2 Treatment Variables

Six months old Jyothi seeds, were subjected to ten mid-storage invigoration treatments. These treatments included control-untreated seeds (T<sub>1</sub>), hydration-dehydration treatment (1l/kg of seeds) (T<sub>2</sub>), ascorbic acid (10g/kg of seeds) (T<sub>3</sub>), calcium chloride (10g/kg of seeds) (T<sub>4</sub>), albizzia leaf powder (2g/kg of seeds) (T<sub>5</sub>), neem cake (5g/kg of seeds) (T<sub>6</sub>), neem leaf powder (2g/kg of seeds) (T<sub>7</sub>), neem oil (2ml/kg of seeds) (T<sub>8</sub>), red chilli powder (1g/kg of seeds) (T<sub>9</sub>) and vayambu leaf powder (2g/kg of seeds) (T<sub>10</sub>).

### 2.3 Environment Variables

The research was carried out at the Regional Agricultural Research Station at Pattambi, Palakkad, which is roughly 60 kilometers north of

Palakkad on the banks of the Bharathapuzha River. This location has a warm, humid tropical climate with relative humidity levels over 70% for the majority of the year. Geographically, the field is located at 10.81°N latitude and 76.19°E longitude.

## 2.4 Seed Quality Analysis

To evaluate seed quality during seed storage, mid-storage treatments were applied to seeds of the Jyothi variety. Important metrics such as germination percentage, seedling vigor index I, mean germination time, and electrical conductivity were calculated.

### 2.4.1 Germination percentage

Germination tests were conducted on rice seeds using a standard method recommended by [13] and [14]. Four replicates of 100 seeds were placed in petri-dishes containing moistened filter papers and kept in a germinator at a temperature of 25°C for two weeks. Observations were recorded daily, and the final count was taken on the 14<sup>th</sup> day. Germination rate (%) was calculated based on the radicle emergence of at least 2 mm .

$$\text{Germination \%} = \frac{\text{Number of seeds germinated}}{\text{Total number of seeds}} \times 100$$

### 2.4.2 Seedling vigour Index I

The seedling viability index -I was calculated according to the formula [15]

$$\begin{aligned} \text{Vigour index I} \\ = \text{Germination (\%)} \times \text{seedling length (cm)} \end{aligned}$$

### 2.4.3 Electrical conductivity

Electrical Conductivity was tested using four replicates of five grams of grains weighed to two decimal places per treatment. After that, the seeds were allowed to soak in 25 milliliters of distilled water and left for 12 hours in an incubator set at 25 ± 10°C. After collecting the seed leachate, distilled water was added to get the volume up to 25 milliliters. The electrical conductivity of the seed leachate was tested using a digital conductivity bridge, and the mean findings were expressed as desi Siemens per meter (dSm<sup>-1</sup>) with a cell constant of 1.0 [16] and [17].

### 2.4.4 Mean germination time

The seeds that were placed on a paper medium to germinate have started sprouting. They were checked daily at the same time, and any seedlings with a radicle protruding 2mm in length were removed from the test. This was continued until all the normal seedlings had germinated to the prescribed size. The mean germination time was then calculated referring to [18,19] and [20].

$$\text{Mean Germination Time (MGT)} = \frac{\sum(n \times d)}{N}$$

where n = number of seeds germinated on each day, d = number of days from the beginning of the test, and N = total number of seeds germinated at the termination of the experiment.

## 2.5 Statistical Analysis

The recorded data was statistically analyzed using the General R-shiny based Analysis Platform Empowered by Statistics (GRAPES), developed by Kerala Agricultural University [21]. The data obtained from seed quality analysis were analyzed using Analysis of Variance (ANOVA) based on the Complete Randomized Design (CRD). The mid-storage invigoration treatments were ranked based on seed quality parameters in seed storage using Duncan's Multiple Range Test (DMRT).

## 3. RESULTS AND DISCUSSION

To ensure efficient seed storage, uniform germination, seedling vigor index, electrical conductivity and mean germination time are the most important elements. By improving the quality of the seed and enhancing its vigor, midstorage invigoration treatments can extend the storage period and promote good seed germination and crop establishment. In this study, we investigated the impact of different midstorage invigoration seed treatments on seed quality parameters for longer storage under ambient conditions of paddy variety Jyothi. The results of the study are discussed below.

### 3.1 Impact of Midstorage Invigoration Seed Treatments on Seed Quality

The analysis of variance for seed quality parameters revealed significant variation, as illustrated in Figs. 1, 2, and Tables 1, 2. The specifics are as follows.

**Table 1. Effect of seed treatment on electrical conductivity of six month old seeds stored under ambient storage condition**

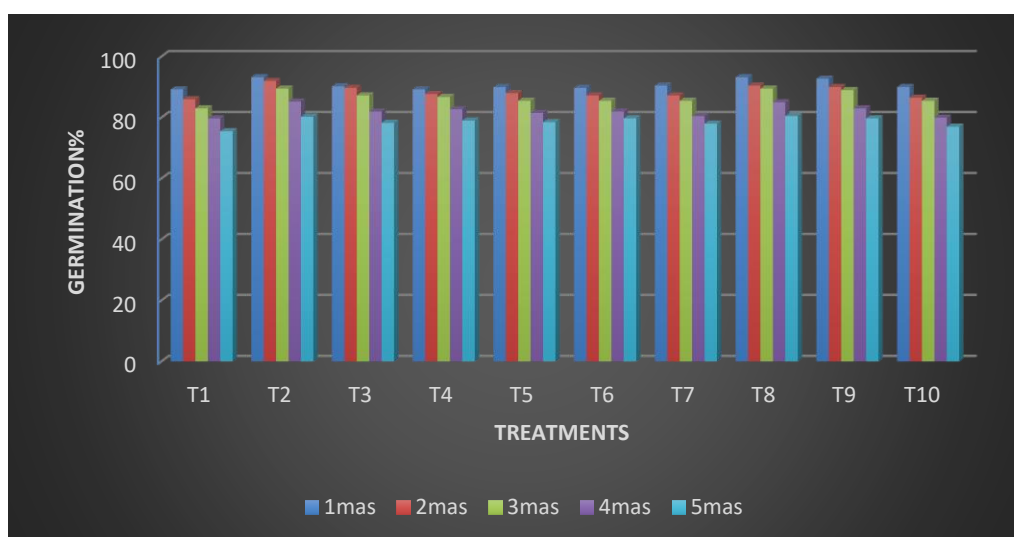
Treatments	1mas	2mas	3mas	4mas	5mas
T <sub>1</sub> :Control	0.791	0.816	0.907	0.997	1.943
T <sub>2</sub> :Hydration - dehydration	0.637	0.692	0.759	0.813	1.012
T <sub>3</sub> :Ascorbic acid	0.702	0.729	0.804	0.880	1.530
T <sub>4</sub> :Calcium chloride	0.702	0.734	0.817	0.900	1.743
T <sub>5</sub> :Albizzia leaf powder	0.780	0.798	0.881	0.910	1.808
T <sub>6</sub> :Neem cake	0.780	0.802	0.900	0.997	1.757
T <sub>7</sub> :Neem leaf powder	0.776	0.805	0.907	1.000	1.833
T <sub>8</sub> :Neem oil	0.778	0.797	0.889	0.977	1.450
T <sub>9</sub> :Redchilli powder	0.789	0.801	0.897	0.993	1.730
T <sub>10</sub> :Vayambu leaf powder	0.788	0.809	0.901	0.990	1.890
<b>C.D(0.05)</b>	0.006	0.004	0.009	0.007	0.051
<b>SE(m)</b>	0.002	0.001	0.003	0.002	0.017

\*1Mas –one month after storage, 2Mas- two months after storage, 3Mas- three months after storage, 4Mas- four months after storage, 5Mas- 5 months after storage

**Table 2. Effect of seed treatment on mean germination time of six month old seeds stored under ambient storage condition**

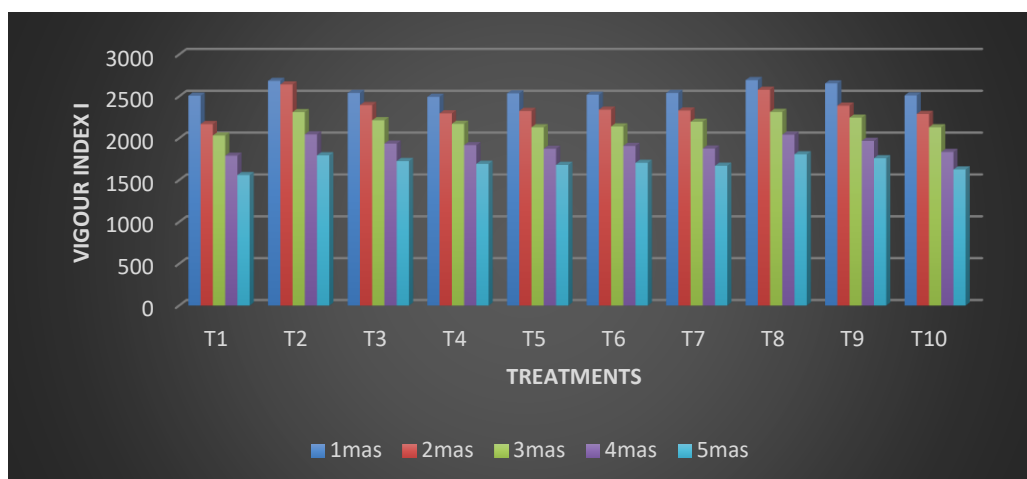
Treatments	1mas	2mas	3mas	4mas	5mas
T <sub>1</sub> :Control	4.37	4.39	4.54	4.69	5.00
T <sub>2</sub> :Hydration - dehydration	4.07	4.14	4.43	4.38	4.53
T <sub>3</sub> :Ascorbic acid	4.12	4.34	4.60	4.54	4.55
T <sub>4</sub> :Calcium chloride	4.15	4.30	4.61	4.60	4.61
T <sub>5</sub> :Albizzia leaf powder	4.12	4.31	4.60	4.56	4.60
T <sub>6</sub> :Neem cake	4.15	4.26	4.62	4.59	4.61
T <sub>7</sub> :Neem leaf powder	4.18	4.34	4.63	4.60	4.63
T <sub>8</sub> :Neem oil	4.06	4.14	4.37	4.40	4.45
T <sub>9</sub> :Redchilli powder	4.11	4.17	4.44	4.39	4.43
T <sub>10</sub> :Vayambu leaf powder	4.28	4.37	4.50	4.53	4.79
<b>C.D(0.05)</b>	0.11	0.084	0.066	0.083	0.133
<b>SE(m)</b>	0.038	0.029	0.023	0.029	0.046

\*1Mas –one month after storage, 2Mas- two months after storage, 3Mas- three months after storage, 4Mas- four months after storage, 5Mas- 5 months after storage



**Fig. 1. Effect of seed treatment on germination percentage of six month old seeds stored under ambient storage condition**

\*Mas – months after storage



**Fig. 2. Effect of seed treatment on seedling vigour index I of six month old seeds stored under ambient storage condition**  
\*Mas – months after storage

### 3.2 Germination Percentage

The results of the study on the impact of seed treatments on germination percentage over time are presented in Fig. 1. From the first month of storage, there were significant differences in germination percentage among the treatments. The germination percentage gradually decreased with increase in storage period. The highest germination percentage after one month of storage was observed in T<sub>8</sub>, which used neem oil at a rate of 2ml/kg (93.25%). T<sub>2</sub>, which used hydration-dehydration treatment at a rate of 1l/kg, and T<sub>9</sub>, which used red chilli powder at a rate of 1g/kg, had a germination percentage of 92.75%. The lowest percentage was recorded in T<sub>1</sub>, which was the control group, with a germination percentage of 89.25%.

Around four months after being stored, the T<sub>1</sub>-control group of seeds were no longer able to maintain 80% germination, unlike the other treated seeds. During the storage period, every other treatment group, except for T<sub>2</sub> (hydration-dehydration treatment 1l/kg) and T<sub>8</sub> (neem oil 2ml/kg treatment), lost minimal germination percentage.

All the mid-storage invigoration treatments were able to significantly improve the germination rate of the treated seeds compared to the control group under normal storage conditions. The treatments were able to maintain the Minimum Seed Certification Standard of 80% germination rate. The neem oil and hydration-dehydration treatments showed the best results over the

other treatments by increasing the storage duration of two months and maintaining minimum germination percentage under normal storage conditions.

Hydration dehydration improved the germination of seeds. This is in accordance with the results of Adhikary and Mandal [22] which states that the act of pre-soaking seeds activates repair mechanisms and metabolic processes that occur during water absorption, resulting in improved germination rates. Neem oil was found to be significantly more effective at keeping the storage insect under control and preserving high seed quality in storage which has been found concurrent with the results of [23]. Dry treatments with red chilli powder 1g/kg was recommended for the improvement of storability of seeds. This was evident with the results of Sengupta et al [24].

### 3.3 Seedling Vigour Index I

Based on the data presented in Fig. 2, it is evident that there were significant differences in the treatments for the various months of storage in terms of the vigour index. The mean vigour index gradually declined after reaching its peak in the first month of storage. In the first month of storage, T<sub>8</sub>-neem oil 2ml/kg (2694) showed a significantly higher vigour index which was comparable to T<sub>2</sub>-hydration dehydration treatment 1l/kg (2684) and T<sub>9</sub>-red chilli powder 1g/kg (2654). The lowest value was found in the control group (2508). Similar trends were observed in the third and fifth months of storage.

Neem oil treatment was found to significantly increase seedling vigour index I. This is similar with the results of Archana et al [25]. T<sub>2</sub>-hydration dehydration treatment resulted in a greater seedling vigour index [26].

### 3.4 Electrical Conductivity

During the storage period, the conductivity gradually increased despite the various treatments applied as represented in Table 1. Seed degradation indicated by seed solute leakage can be measured by electrical conductivity. It has been found that biological membranes control the flow of substances into and out of cells, suggesting that they are crucial for the viability and vigor of seeds. Under ambient storage conditions, it was observed that in the first month, T<sub>1</sub>- control had the highest electrical conductivity of seed leachate (0.791 dSm<sup>-1</sup>), while the least EC was recorded in T<sub>2</sub>-hydration dehydration treatment (0.637 dSm<sup>-1</sup>) and T<sub>3</sub>- ascorbic acid (0.702 dSm<sup>-1</sup>). At the end of storage, the highest electrical conductivity was recorded in T<sub>1</sub>- control (1.943 dSm<sup>-1</sup>), which is consistent with the findings of Kavita, Yogeesh [27] and Al Maskri et al. [28]. As electrical conductivity points to seed solute leakage, lower EC values are correlated with the good seed quality parameters. In the last month of storage lower electrical conductivity values were ascertained in the seeds treated with T<sub>2</sub>-hydration dehydration treatment (1.012 dSm<sup>-1</sup>) and T<sub>8</sub>-neem oil 2ml/kg (1.450 dSm<sup>-1</sup>) brought about prolonged seed storage.

### 3.5 Mean Germination Time

The average germination time is a crucial factor in determining the quality of any seed lot, as it reflects the speed of germination. According to Zhang et al [29], the mean germination time can be used to evaluate seed vigor. In this study, the mean germination time increased during the storage period, as shown in Table 2. Initially, T<sub>8</sub>-neem oil and T<sub>2</sub>-hydration dehydration treatment had low mean germination times (4.06 and 4.07 days, respectively), but they increased to 5 days in the control group (T<sub>1</sub>). However, at the end of storage, seeds treated with T<sub>9</sub>-red chilli powder, T<sub>8</sub>-neem oil, and T<sub>2</sub>-hydration dehydration treatment had mean germination times of 4.43, 4.45, and 4.53 days, respectively. These treatments were superior to all other treatments at the end of the storage period. The highest mean germination time of 5 days was observed in the control group (T<sub>1</sub>). These findings are

consistent with those of Garoma et al. [30]. The reduced lag phase is associated with an increase in the mean germination time [31-33].

## 4. CONCLUSION

The study on Jyothi paddy seeds subjected to various mid-storage invigoration treatments showed significant impacts on seed quality parameters. Out of all the treatments, neem oil, hydration-dehydration, and red chili powder yielded the most favorable outcomes for enhanced seed longevity. The research findings imply that farmers could potentially augment seed quality and prolong seed storage by employing eco-friendly and economically viable midstorage invigoration seed treatments. Farmers have easy access to three useful resources: neem oil, hydration-dehydration techniques, and red chili powder. By incorporating these readily available solutions into their seed storage practices, farmers can extend the seed storage duration and make use of the paddy seeds for next crop season with maintaining the required minimum germination potential.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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